

This Listing of Claims will replace all prior versions, and listings, of claims in this application:

Listing of Claims:

1. (currently amended): An isolated or purified nucleic acid molecule comprising a nucleotide sequence at least 95% identical to SEQ ID NO:1 and which codes for a pyruvate carboxylase enzyme desensitized to feedback inhibition by aspartic acid, wherein said pyruvate carboxylase enzyme contains at least one mutation to SEQ ID NO:19, ~~which desensitizes said pyruvate carboxylase enzyme to feedback inhibition by aspartic acid~~, wherein said mutation is selected from the group consisting of:

- a) methionine at position 1 is replaced with a valine,
- b) glutamic acid at position 153 is replaced with an aspartic acid,
- c) alanine at position 182 is replaced with a serine,
- d) alanine at position 206 is replaced with a serine,
- e) histidine at position 227 is replaced with an arginine,
- f) alanine at position 452 is replaced with a glycine, and
- g) aspartic acid at position 1120 is replaced with a glutamic acid.

2. (currently amended): An isolated or purified nucleic acid molecule comprising a nucleotide sequence selected from the group consisting of:

- a) the nucleotide sequence encoding amino acids 1 to 1157 of SEQ ID NO:2;
- b) a nucleotide sequence encoding the amino acid sequence encoded by the DNA

plasmid encoding feedback resistant pyruvate carboxylase enzyme, said plasmid contained in

Deposit Number NRRL B-30293; and

c) a nucleotide sequence completely complementary to any of the nucleotide sequences (a) or (b).

3. (previously presented): The nucleic acid molecule of claim 2, comprising the nucleotide sequence of SEQ ID NO:1.

4. (cancelled).

5. (previously presented): A vector comprising:

a) the nucleic acid molecule of claim 1 or 2; and

b) at least one marker gene.

6. (previously presented): The vector of claim 5, further comprising a functional *Corynebacterium* replication origin.

7. (previously presented): A method for producing a host cell comprising introducing the vector of claim 5 into a host cell.

8. (previously presented): A host cell comprising the vector of claim 5.

9. (withdrawn): A method of producing an amino acid, comprising:

a) culturing the host cell of claim 8, in a suitable media; and

b) separating said amino acid from said medium.

10. (withdrawn): The method of claim 9, wherein said amino acid is selected from the group consisting of: L-lysine, L-threonine, L-methionine, L-isoleucine, L-glutamic acid, L-arginine and L-proline.

11. (withdrawn): The method of claim 10, wherein said amino acid is L-lysine.
12. (previously presented): A method for replacement of a wild-type pyruvate carboxylase gene, with a feedback resistant pyruvate carboxylase gene, in a *Corynebacterium glutamicum* host cell comprising the steps of:
 - a) replacing a genomic copy of said wild-type pyruvate carboxylase gene with a selectable marker gene through homologous recombination to form a first recombinant strain; and
 - b) replacing said selectable marker gene of step (a) in said first recombinant strain, with said feedback resistant pyruvate carboxylase gene through homologous recombination to form a second recombinant strain;wherein said homologous recombination in steps (a) and (b) occurs between said host cell and the vector of claim 5.
13. (previously presented): A host cell produced by the method of claim 12.
14. (withdrawn): A method of producing an amino acid, comprising:
 - a) culturing the host cell of claim 13 in a suitable medium; and
 - b) separating said amino acid from said medium.
15. (withdrawn): The method of claim 14, wherein said amino acid is selected from the group consisting of: L-lysine, L-threonine, L-methionine, L-isoleucine, L-glutamic acid, L-arginine and L-proline.
16. (withdrawn): The method of claim 15, wherein said amino acid is L-lysine.

17. (withdrawn): An isolated or purified polypeptide comprising the amino acid sequence of the polypeptide encoded by the DNA plasmid encoding pyruvate carboxylase contained in Deposit Number NRRL B-11474, the amino acid sequence of SEQ ID NO:2 or the amino acid sequence of SEQ ID NO:4.

18. (withdrawn): An isolated or purified polypeptide comprising an amino acid sequence selected from the group consisting of: SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16 and SEQ ID NO:18.

19. (currently amended): An isolated or purified nucleic acid molecule comprising a nucleotide sequence at least 95% identical to SEQ ID NO:1 and which codes for a pyruvate carboxylase enzyme desensitized to feedback inhibition by aspartic acid, the nucleic acid molecule comprising a nucleotide sequence encoding a polypeptide comprising ~~an~~ the amino acid ~~sequence~~ sequences set forth in ~~selected from the group consisting of:~~ SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16 and SEQ ID NO:18.

20. (currently amended): The nucleic acid molecule of claim 19, wherein said nucleic acid molecule comprises a the nucleotide ~~sequence~~ sequences set forth in ~~selected from the group consisting of:~~ SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11 SEQ ID NO:13, SEQ ID NO:15 and SEQ ID NO:17.

Claims 21-23. (cancelled).

24. (new): An isolated or purified nucleic acid molecule comprising a nucleic acid sequence encoding a pyruvate carboxylase enzyme desensitized to feedback inhibition by aspartic acid,

said enzyme having an amino acid sequence that differs from SEQ ID NO: 19 by at least one mutation, said at least one mutation selected from the group consisting of:

- a) methionine at position 1 is replaced with a valine,
- b) glutamic acid at position 153 is replaced with an aspartic acid,
- c) alanine at position 182 is replaced with a serine,
- d) alanine at position 206 is replaced with a serine,
- e) histidine at position 227 is replaced with an arginine,
- f) alanine at position 452 is replaced with a glycine, and
- g) aspartic acid at position 1120 is replaced with a glutamic acid.

25. (new): An isolated or purified nucleic acid molecule comprising a nucleotide sequence at least 95% identical to SEQ ID NO:1 and which codes for a pyruvate carboxylase enzyme desensitized to feedback inhibition by aspartic acid, wherein said pyruvate carboxylase enzyme contains at least seven mutations to SEQ ID NO:19, wherein said at least seven mutations to SEQ ID NO:19 include:

- a) methionine at position 1 is replaced with a valine,
- b) glutamic acid at position 153 is replaced with an aspartic acid,
- c) alanine at position 182 is replaced with a serine,
- d) alanine at position 206 is replaced with a serine,
- e) histidine at position 227 is replaced with an arginine,
- f) alanine at position 452 is replaced with a glycine, and
- g) aspartic acid at position 1120 is replaced with a glutamic acid.

26. (new): A vector comprising:

- (a) the nucleic acid molecule of claim 24 or 25; and
- (b) at least one marker gene.

27. (new): The vector of claim 26, further comprising a functional *Corynebacterium* replication origin.

28. (new): A method for producing a host cell comprising introducing the vector of claim 26 into a host cell.

29. (new): A host cell comprising the vector of claim 26.

30. (new): A method for replacement of a wild-type pyruvate carboxylase gene, with a feedback resistant pyruvate carboxylase gene, in a *Corynebacterium glutamicum* host cell comprising the steps of:

- (a) replacing a genomic copy of said wild-type pyruvate carboxylase gene with a selectable marker gene through homologous recombination to form a first recombinant strain; and

- (b) replacing said selectable marker gene of step (a) in said first recombinant strain, with said feedback resistant pyruvate carboxylase gene through homologous recombination to form a second recombinant strain;

wherein said homologous recombination in steps (a) and (b) occurs between said host cell and the vector of claim 26.

31. (new): A host cell produced by the method of claim 30.